

In the Claims

Please cancel claims 11, 12, 19-26, 28-33, 35-41, 43-48, 50-52, 54-62, 64-68, 72, 74-83, 85-89, 91-95, 98-106, 108-115 and 117-133. Please amend claims 49, 53 and 69. Claims 1-10, 13-18, 27, 34, 42, 49, 53, 63, 69-71, 73, 84, 90, 96, 97, 107 and 116 are now pending.

1. (Original) A method of making a polysaccharide over-producing bacterium comprising introducing into a bacterium an *ica* nucleic acid operably linked to an *ica* regulatory nucleic acid,
wherein the *ica* regulatory nucleic acid comprises
 - (a) nucleic acid molecules which hybridize under stringent conditions to a nucleic acid molecule having a sequence of SEQ ID NO:2, have an addition, deletion or substitution in a region between and including nucleotides 9 and 43 of SEQ ID NO:2, and that enhance production of a polysaccharide from an *ica* locus, and
 - (b) complements thereof.
2. (Original) The method of claim 1, wherein the bacterium is a *Staphylococcus* bacterium.
3. (Original) The method of claim 2, wherein the *Staphylococcus* bacterium is selected from the group consisting of *Staphylococcus epidermidis*, *Staphylococcus aureus*, *Staphylococcus capitis*, *Staphylococcus caprae*, *Staphylococcus hemolyticus*, *Staphylococcus auricularis*, *Staphylococcus intermedius*, *Staphylococcus lugdunensis*, *Staphylococcus pasteurii*, and *Staphylococcus piscifermentans*.
4. (Original) The method of claim 1, further comprising measuring polysaccharide production from the bacterium, wherein a high level of polysaccharide production is indicative of a polysaccharide over-producing bacterium.
5. (Original) The method of claim 1, wherein the *ica* regulatory nucleic acid comprises the nucleotide sequence of SEQ ID NO:1.

6. (Original) The method of claim 1, wherein the *ica* regulatory nucleic acid comprises the nucleotide sequence between and including nucleotides 9 and 38 of SEQ ID NO:1.
7. (Original) The method of claim 1, wherein the *ica* regulatory nucleic acid comprises a deletion, addition or substitution in the region between and including nucleotides 24 and 28 of SEQ ID NO:2.
8. (Original) The method of claim 1, wherein the *ica* regulatory nucleic acid comprises a five nucleotide non-wildtype substitution between and including nucleotides 24 and 28 of SEQ ID NO:2.
9. (Original) The method of claim 8, wherein the five nucleotide non-wildtype substitution has a sequence of ATAAA.
10. (Original) A method of making a polysaccharide over-producing bacterium comprising introducing into a bacterium an *ica* nucleic acid operably linked to an *ica* regulatory nucleic acid, wherein the *ica* regulatory nucleic acid comprises a mutant *icaR* nucleic acid, and measuring polysaccharide production from the bacterium, wherein a high level of polysaccharide production is indicative of a polysaccharide over-producing bacterium.
- 11-12. (Cancelled)
13. (Original) The method of claim 10, wherein the mutant *icaR* nucleic acid does not encode a wildtype IcaR protein.
14. (Original) The method of claim 10, wherein the mutant *icaR* nucleic acid comprises a frameshift mutation relative to a wildtype *icaR* nucleic acid.
15. (Original) The method of claim 10, wherein the mutant *icaR* nucleic acid encodes a truncated IcaR protein.

16. (Original) The method of claim 10, wherein the mutant *icaR* nucleic acid encodes a mutant IcaR protein that binds to a target less efficiently than wildtype IcaR protein.
17. (Original) The method of claim 10, wherein the polysaccharide is PNAG.
18. (Original) A method of making a polysaccharide over-producing bacterium comprising recombinantly down-regulating wildtype IcaR protein production, and selecting a polysaccharide over-producing bacterium.
- 19-26. (Cancelled)
27. (Original) A method of making a polysaccharide over-producing bacterium comprising recombinantly altering the TATTT nucleotide sequence in the *ica* promoter region.
- 28-33. (Cancelled)
34. (Original) A recombinant polysaccharide over-producing bacterium comprising an *ica* nucleic acid operably linked to an *ica* regulatory nucleic acid,
wherein the *ica* regulatory nucleic acid comprises
(a) nucleic acid molecules which hybridize under stringent conditions to a nucleic acid molecule having a sequence of SEQ ID NO:2, have an addition, deletion or substitution in a region between and including nucleotides 9 and 43 of SEQ ID NO:2, and that enhance production of a polysaccharide from an *ica* locus, and
(b) complements thereof.
wherein the bacterium is not MN8m.
- 35-41. (Cancelled)
42. (Original) A recombinant polysaccharide over-producing bacterium comprising a mutant *icaR* nucleic acid.

43-48. (Cancelled)

49. (Currently Amended) A method of producing a bacterial polysaccharide comprising culturing the polysaccharide over-producing bacterium of claim 34[[-47 or 48]] in a growth medium, and
harvesting the bacterial polysaccharide from the culture.

50-52. (Cancelled)

53. (Currently Amended) A method of producing an antibody to a bacterial polysaccharide comprising
isolating a bacterial polysaccharide from the polysaccharide over-producing bacterium of claim 34[[-47 or 48]],
administering to a subject the isolated bacterial polysaccharide in an amount effective to produce an antibody, and
harvesting antibody from the subject.

54-62. (Cancelled)

63. (Original) An isolated nucleic acid molecule, comprising
(a) nucleic acid molecules which hybridize under stringent conditions to a nucleic acid molecule having a sequence of SEQ ID NO:2, have an addition, deletion or substitution in a region between and including nucleotides 9 and 43 of SEQ ID NO:2, and that enhance production of a polysaccharide from an *ica* locus, and
(b) complements thereof.

64-68. (Cancelled)

69. (Currently Amended) An expression vector comprising the isolated nucleic acid molecule of claim 63[[-67 or 68]], operably linked to an *ica* nucleic acid.

70. (Original) A host cell transformed or transfected with the expression vector of claim 69.

71. (Original) An isolated nucleic acid molecule selected from the group consisting of
(a) a fragment of a nucleic acid molecule having a sequence of SEQ ID NO:1, and
(b) complements of (a),

wherein the fragment spans a MN8m mutation and enhances production of a polysaccharide from an *ica* locus when operably linked to an *ica* nucleic acid.

72. (Cancelled)

73. (Original) A method for identifying an isolated binding agent, comprising
contacting a first nucleic acid molecule having the sequence of SEQ ID NO:2 or a functionally equivalent fragment thereof with a candidate molecule and determining whether the candidate molecule binds to the first nucleic acid molecule, and

contacting a second nucleic acid molecule having the sequence of SEQ ID NO:1 or a functionally equivalent fragment thereof with the candidate molecule and determining whether the candidate molecule binds to the second nucleic acid molecule,

wherein a candidate molecule that binds to either the first or the second nucleic acid molecule but not both is indicative of an isolated binding agent.

74-83. (Cancelled)

84. (Original) A method of identifying an *ica* promoter sequence associated with polysaccharide overproduction comprising

detecting a nucleic acid molecule having a sequence alteration from wildtype in a region between and including nucleotides 9 and 43 of SEQ ID NO:2.

85-89. (Cancelled)

90. (Original) A method for identifying an *ica* regulatory nucleic acid molecule that enhances polysaccharide production comprising

altering a nucleic acid molecule having a sequence of SEQ ID NO:2, and
determining a level of reporter production by a bacterium that comprises the altered
nucleic acid molecule operably linked to reporter nucleic acid,

wherein a higher than wildtype level of reporter protein production is indicative of an *ica*
regulatory nucleic acid molecule that enhances polysaccharide production.

91-95. (Cancelled)

96. (Original) A composition comprising
an isolated binding agent that binds to a nucleic acid having a sequence of SEQ ID NO:1
with greater affinity than to SEQ ID NO:2.

97. (Original) A composition comprising
an isolated binding agent that binds to a nucleic acid having a sequence of SEQ ID NO:2
with greater affinity than to SEQ ID NO:1.

98-106. (Cancelled)

107. (Original) A method of over-producing a protein in a bacterium comprising
introducing into a bacterium a nucleic acid operably linked to an *ica* regulatory nucleic
acid,

wherein the *ica* regulatory nucleic acid comprises

(a) nucleic acid molecules which hybridize under stringent conditions to a nucleic
acid molecule having a sequence of SEQ ID NO:2, have an addition, deletion or substitution in a
region between and including nucleotides 9 and 43 of SEQ ID NO:2, and that enhance
production of a polysaccharide from an *ica* locus, and

(b) complements thereof, and

wherein the nucleic acid encodes a protein to be over-produced.

108-115. (Cancelled)

116. (Original) A method of over-producing a protein in a bacterium comprising introducing into a bacterium a nucleic acid operably linked to an *ica* regulatory nucleic acid, wherein the *ica* regulatory nucleic acid comprises a mutant *icaR* nucleic acid, wherein the nucleic acid encodes a protein to be over-produced.

117-133. (Cancelled)